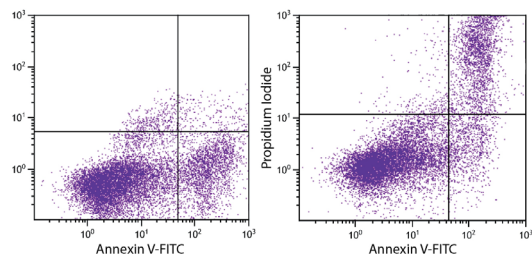


ApoScreen® Annexin V Apoptosis Kit-FITC

Cat. No.	Kit Format	Size
10010-02	Fluorescein (FITC) Conjugated Annexin V	100 tests



Day old BALB/c mouse splenocytes were stained with ApoScreen® Annexin V Apoptosis Kit-FITC (SB Cat. No. 10010-02).

Description

ApoScreen® Annexin V Apoptosis Kit-FITC (SB Cat. No. 10010-02) employs a fluorescein-conjugated Annexin V (Annexin V-FITC) in concert with propidium iodide (PI) to evaluate subpopulations of cells undergoing apoptosis. During the early stages of apoptosis, cells begin to display phosphatidylserine (PS) on the outer cell membrane where it is readily detectable by staining the cells with Annexin V-FITC. As the plasma membrane becomes increasingly permeable during the later stages of apoptosis, PI can readily move across the cell membrane and bind to cellular DNA, providing a means for identifying those cells that have lost membrane integrity through mechanisms including necrosis. When cells are double stained with Annexin V-FITC and PI, three different cell populations may be observed - (i) live cells that do not stain with either Annexin V-FITC or PI; (ii) necrotic cells that stain with both reagents; and (iii) apoptotic cells that stain with Annexin V-FITC only. Analysis may be performed on any flow cytometer equipped with a single laser using excitation at 488 nm. Alternatively, the cells can be examined with a fluorescence microscope equipped with filters for discrimination of FITC and rhodamine/PI.

Applications

FC – Quality tested ^{1-7, 9-34}
 ICC – Reported in literature ^{6,8,35}

Kit Components & Storage and Handling

- Annexin V-FITC is supplied as 100 tests in 1.0 mL of Annexin V Binding Buffer. Store at 2-8°C.
- Propidium Iodide (PI) is supplied as 100 tests in 1.0 mL of Annexin V Binding Buffer. Store at 2-8°C.
- Annexin V Binding Buffer 10X is supplied as 7 x 1.5 mL. Store at 2-8°C.
- Protect FITC conjugated and PI reagents from light. Reagents are stable for the period shown on the label if stored as directed.

Working Dilutions

Flow Cytometry	Annexin V-FITC	10 µL/10 ⁶ cells
	Propidium Iodide (PI)	10 µL/10 ⁶ cells
	Annexin Binding Buffer 10X	Dilute 1 part 10X buffer with 9 parts dH ₂ O just prior to use
	For flow cytometry, the suggested use of these reagents is in a final volume of 100 µL of Annexin Binding Buffer 1X	

Other Applications Since applications vary, you should determine the optimum working dilution for the product that is appropriate for your specific need.

Suggested Staining Protocol

- Wash cells twice in cold PBS
- Resuspend cells in cold 1X Annexin Binding Buffer to a concentration of 1 x 10⁶ cells/mL
- Add 100 µL of cell suspension (10⁶ cells) to 5 mL polystyrene round bottom tubes
- Add 10 µL of conjugated Annexin V-FITC
- Gently vortex each tube and incubate for 15 minutes at 2-8°C and protected from light
- Without washing, add ~380 µL of cold 1X Annexin Binding Buffer to each tube
- Add 10 µL of Propidium Iodide (PI)
- Analyze by flow cytometry immediately

For Research Use Only. Not for Diagnostic or Therapeutic Use.

Warning

Some reagents contain sodium azide. Please refer to product specific (M)SDS.

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